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1: Biotechnology (N Y). 1993 Oct;11(10):1138-43. 

# Use of peptide libraries to map the substrate specificity of a peptide-modifying enzyme: a 13 residue consensus peptide specifies biotinylation in Escherichia coli.

**Schatz PJ.**

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I describe a technique for screening peptide libraries of over 10(9) independent clones for substrates of peptide-modifying enzymes. The peptides, linked to their genetic material by the lac repressor, are exposed to the enzyme and then screened by affinity purification on a receptor specific for the modified product. The enzyme characterized, E. coli biotin holoenzyme synthetase, normally adds biotin to a specific lysine residue in complex protein domains. The 13 residue substrate identified by this library screening approach is much smaller than the 75 amino acid required sequence of the natural substrate, and can function at either end of a fusion protein. The sequence is quite distinct at some positions from that region of the natural substrates, presumably because the peptides have to mimic the folded structure formed by the natural substrate. This technique should be useful for mapping the substrate specificity of a variety of peptide-modifying enzymes. In addition, small peptide substrates that are enzymatically biotinylated at a single site should be useful for a variety of purposes in labeling, purification, detection, and immobilization of proteins.

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